about a paradigm shift in our understanding of its role in health and disease and has substantial consequences for microbiome design and human health issues.

EDNA-Water, using deep sequencing and bioinformatics approach for water-borne plant virus detection
L. PENA ZUNIGA, J. Daniels, A. Espindola, F. Ochoa Corona, Oklahoma State University, Stillwater, OK, USA

Agriculture is adversely affected by viral infections. Water-borne plant viruses: *Potexvirus, Tobamovirus* and *Tombusvirus*, have been reported worldwide and a number of them occur in the U.S. Viral infections in irrigated fields or hydroponic crops are not detected until symptoms are evident. Single-virus serological and molecular assays are used for detection. Third-generation sequencing (TGS) offers a broad detection capability to identify multiple unique Nucleic Acid signatures in metagenomic data from a single sample. Electronic probe Diagnostic Nucleic acid Analysis (EDNA) was developed to detect water-borne plant viruses in simulated mock metagenomic sequence databases (MSDs), assembled with reference positive controls and host genome sequences. MSDs were generated using MetaSim and configured to simulate Illumina average read length and error rates. Virus E-probes ranging 20-60 nucleotides lengths were designed using EDNA MiProbe software. MSDs containing host and the target viral genome were used as positive controls. MSDs mimicking single and multiple infections were also generated. EDNA E-probes proved to be a relatively rapid diagnostic method for specific and discrete targeted viruses in water if compared with other TGS bioinformatics analyses. This provides the framework for a new sequence-based diagnosis system to make disease management decisions.

Surprises learned from plant immunity –challenges and opportunities for crop protection
Y. JIA (1), H. Zhao (2), X. Wang (3), (1) USDA ARS, Dale Bumpers National Rice Research Center, Stuttgart, AR, USA; (2) USDA ARS, 130, AR, USA; (3) UA/USDA, Stuttgart, AR, USA

Plant species have coevolved with their pathogens and both have survived under changing environments. The molecular mechanisms of disease resistance have been a focus of intense research over the last two decades. From the simplest model of elicitor and receptor interaction, to the complex active and passive defense responses learned from the plant kingdom, research has illustrated that plants have evolved remarkable multifaceted and sophisticated defense systems. These defense systems include localized and systemic responses all of which are governed by major and minor resistance (*R*) genes. For example, rice *R* genes that prevent infections by the fungus *Magnaporthe oryzae* were found among all rice chromosomes. Some blast *R* genes and genes controlling plant productivity are co-localized on recombination suppressed chromosomal regions of the rice genome. As a result, some robust *R* genes were lost and not effectively utilized due to domestication and extensive crop improvement efforts. Continued investigation of molecular mechanisms of plant immunity and cross-talk among genes involved in productivity can be enhanced by effective utilization of recent cutting edge technology like CRISPR-Cas 9 gene editing for crop protection. Contemporary knowledge of plant immunity and development of utilization strategies for crop improvement will be presented.

Control of cereal pathogens in the light of resistance development in Europe
A. Rehfus, A. Huf, R. J. Bryson, D. Strobel, G. STAMMLER, BASF SE, Limburgerhof, GERMANY

Fungal pathogens in cereals have a significant impact on yield and quality. In wheat, *Zymoseptoria tritici, Blumeria graminis* f.sp. *tritici, Puccinia triticina*, and *Fusarium* spp. are important European pathogens, while in barley *Pyrenophora teres, Ramularia collo-cygni and Rhynchosporium commune* play a key role. Fungicides for disease control include QoIs, SBIs and SDHIs. In countries with intensive QoI use, QoI resistance conferred by the G143A exchange in the *cyt b* has now developed in most important cereal pathogens, except for rusts, *R. commune* and *P. teres*. SBIs have been used for more than 30 years to control cereal diseases. Mutations in the *cyp51* gene are responsible for sensitivity changes with the most advanced evolution of *cyp51* taking place in *Z. tritici*, with many different haplotypes. Additionally, *cyp51* overexpression and enhanced efflux have also been detected as resistance mechanisms. An analysis of several hundred *Z. tritici* isolates from across Europe show a different distribution of *cyp51* haplotypes and *cyp51* overexpression over Europe. Despite this, SBIs are still very important tools in cereal disease control, as well as for resistance management in partnership with the highly effective SDHIs. SDHI resistance has been detected for *P. teres* and *R. collo-cygni*. Several mutations in the *sdh* genes cause resistance and form a complex situation in these two pathogens. *sdh* mutations have also been found in field isolates of *Z. tritici*, but the mutation pattern and dynamic is highly different to the barley pathogens. Knowledge on fitness costs by (multiple) resistance and the effects of management strategies might help in the development of sustainable disease control strategies including also non-chemical approaches.

New approaches to detection: Canine surveillance of high risk pathogens
T. R. GOTTWALD (1), G. Poole (2), G. Mccollum (2), W. Luo PhD (3), F. Louws (4), (1) USDA-ARS, Ft Pierce, FL, USA; (2) USDA, ARS, Fort Pierce, FL, USA; (3) USDA ARS & NCSU, Ft Pierce, FL, USA; (4) North Carolina State University, Raleigh, NC, USA

The underpinning for control of exotic diseases such as citrus Huanglongbing (HLB) caused by Candidatus *Liberibacter asiaticus* (CLas) and Plum Pox Virus (PPV) is early detection/response when incidence is low. Unfortunately, these pathogens can remain asymptomatic and subclinical to PCR detection for months yet can act as inoculum sources. Twenty canines were trained for early detection of HLB and two for PPV. Ten canines were each tested against 1000 trees in replicated randomized field trials with varying HLB-incidence, which resulted in 99.16% overall detection accuracy with very few false negatives or positives. Canines also detected infected trees exclusively from 5-gm feeder root samples. In a time-course experiment, canines detected infections within 2–3 weeks of vector transmission, whereas inoculated trees were not PCR-positive for CLas until at least 3–12 mos. post inoculation. In citrus and prunus field trials, canines trot along the rows with an average interrogation time of ~2–10 trees/s; faster than any other detection method. Canines were also effectively utilized for detection of infected trees in residential areas. This confirms that canines are a very early, accurate and sensitive detection methodology. Canines are able to detect the pathogen in trees with subclinical infection, i.e., before symptom expression and considerably prior to the ability of PCR detection.

Identification of a hypervirulent pathotype of *Rice yellow mottle virus*: A threat to genetic resistance deployment in West-Central Africa
E. HÉBRARD (1), A. Pinel-Galzi (1), A. Oludare (2), N. Poulicard (1), J. Aribi (1), S. Fabre (1), S. Issaka (3), C. Mariac (4), A. Dereeper (1), L. Albar (4), D. Silue (2), D. J. Fargette (1), (1) IRD, Cirad, Univ Montpellier, IPME, Montpellier, FRANCE; (2) AfricaRice, Bouaké, IVORY COAST; (3) FSAE, Université de Tillabéri, Tillabéri, NIGER; (4) IRD, Univ Montpellier, DIADE, Montpellier, FRANCE

Rice yellow mottle virus (RYMV) is a major biotic constraint on rice cultivation in Africa, causing high losses to rice production. Several sources of varietal high resistance are available but the emergence of virulent pathotypes that are able to overcome one or two resistance alleles can sometimes occur. Both resistance spectra and viral adaptability have to be taken into account to develop sustainable rice breeding strategies against RYMV. In this study, we assessed the adaptability of 20 viral isolates representative of the RYMV genetic and pathogenic diversity in Africa against high resistant rice

accessions. Our results revealed a hypervirulent pathotype, named thereafter pathotype T', that is able to overcome all known sources of high resistance. This pathotype, which is spatially localized in West-Central Africa, appears to be more abundant than previously suspected. To better understand the adaptive processes of pathotype T', molecular determinants of resistance-breakdown were identified via Sanger sequencing and were validated through directed mutagenesis of an infectious clone. These analyses confirmed the key role of convergent non-synonymous substitutions in the central part of the VPg to overcome *RYMV1*-mediated resistance. In addition, deep-sequencing analyses revealed that virulence mutations present in a small proportion of the virus population can be sufficient for resistance-breakdown. Considering the spatial distribution of RYMV strains in Africa and their ability to overcome the RYMV resistance genes and alleles, we established a resistance-breaking risk map to optimize strategies for the deployment of sustainable and resistant rice lines in Africa.

Linking molecules with morphology in the -Omics age: Computational taxonomy pipelines for nematodes and other microbial metazoa
H. BIK, University of California, Riverside, Riverside, CA, USA

Microbial metazoa (organisms <1mm, including nematodes, tardigrades, platyhelminthes, other "minor" metazoan phyla, and eggs/larval stages of larger species) are abundant and ubiquitous in soil/sediment habitats, performing key functions such as nutrient cycling and sediment stability in marine and terrestrial ecosystems. Yet, their unexplored diversity represents one of the major challenges in biology and currently limits our capacity to understand, mitigate and remediate the consequences of environmental change. Microbial metazoa have a strong history of morphological taxonomy (formal species descriptions, specimen drawings, monographs, etc.), but most of this information is offline and thus effectively inaccessible for modern day -Omics studies. In addition, DNA barcoding databases and genome collections lag far behind other groups of microbial organisms such as bacteria, archaea, fungi, and single-celled protists. This sparsity of computational resources severely limits the ecological and evolutionary insights that can be gained from high-throughput sequencing approaches focused on microbial metazoa. Here, I will discuss recent efforts to improve molecular databases and expand bioinformatics pipelines for -Omic studies of "neglected" phyla such as nematodes, focusing on tool development as well as community building efforts.

Surveillance for plant pests using meta-barcoding and LAMP techniques
N. BOONHAM (1,2), T. Wood (3), J. Hodgetts (1), I. Adams (1), R. Caiazzo (3), S. Franco Ortega (4), R. Glover (1), M. Andreou (5), (1) Fera Science Ltd, York, UNITED KINGDOM; (2) Newcastle University, Newcastle upon Tyne, UNITED KINGDOM; (3) National Institute of Agricultural Botany, Cambridge, UNITED KINGDOM; (4) Agroinnova - University of Torino, Grugliasco, Torino, ITALY; (5) Optisense Ltd, Horsham, UNITED KINGDOM

The overall aim of the EMPHASIS project is to provide integrated response measures to predict, prevent and protect European agriculture, forestry sectors, and natural ecosystems from native and alien pest threats. The effectiveness of these solutions will be assessed, validated and promoted through co-innovative research and demonstration in line with end user needs and capacities. The talk will focus on the development of targeted and non-targeted methods for the detection and surveillance of quarantine pathogens and pests. LAMP tests have been developed for a range of targets in arable (including wheat, rice and potatoes), horticultural crops (including basil and lettuce) as well as diseases impacting ornamental plants and the environment (e.g. *Xylella fastidiosa*). The work aims to develop and validate to international standards (EPPO) a large range of LAMP assays for a number of different end users in disease diagnosis, seed testing and environmental monitoring. The assays are developed to run on the Genie suite of instruments (Optigene) and the platform has been further developed to facilitate usage, including the development of on-screen protocols, training, and formatting of kits to enhance the end-user experience. Non-targeted detection has focused on the development of meta-barcoding approaches for surveillance of airborne fungal spores and insect pests following trapping. The aim is to profile the species present in the traps and provide broad spectrum data on the presence of quarantine and other pathogens moving in a particular location. We are focusing this surveillance work on pathogens/pests of interest to farmers and regulators to facilitate the implementation of early control measures in each sector.

The mechanism of xylose-dependent expression of *hrp* genes in a rice pathogen *Xanthomonas oryzae* pv. *oryzae*
Y. IKAWA, S. Tsuge PhD, Kyoto Prefectural University, Sakyo-Ku, Kyoto, JAPAN

hrp genes encoding components of the type III secretion system are essential for virulence of *Xanthomonas oryzae* pv. *oryzae* (Xoo), the bacterial pathogen of leaf blight of rice. Expression of *hrp* genes is regulated by two key regulators, HrpG and HrpX. Previously, we reported the importance of xylose in *hrp* gene expression in the bacterium, and that, without xylose, the accumulation of a *hrp* regulator, HrpX, not the expression of the protein, is decreased. Here, to clarify the mechanism of quantitative regulation of HrpX, we conducted random transposon mutagenesis, and found that a mutant with a transposon in a LacI-type transcriptional regulator XylR showed high *hrp* gene expression even under the xylose-free medium. The *xylR* deletion mutant showed high HrpX accumulation and *hrp* gene expression in the medium without xylose, similarly to the wild type incubated in the xylose-containing medium. XylR is known to negatively regulate xylan/xylose metabolism-related genes, including a xylose isomerase gene *xylA2*, in other bacteria. We confirmed the involvement of Xoo XylR in the negative regulation of *xylA2* and, furthermore, the binding of the protein to the upstream region of the *xylA2*. These results suggest that, in the presence of xylose, inactivation of XylR activates xylan/xylose utilization and, simultaneously, of *hrp* gene expression through the higher accumulation of HrpX in Xoo. The mechanism of the XylR-dependent quantitative regulation of HrpX will be discussed.

Quality assurance, validation of tests and collections in plant pest diagnostics: Approaches and experience in the EPPO region
F. PETTER, M. McMullen, B. Giovani, European and Mediterranean Plant Protection Organization, Paris, FRANCE

EPPO is an intergovernmental organization responsible for European cooperation in plant health. The EPPO diagnostic programme started in 1998 with the preparation of pest specific diagnostic protocols. Development of quality management systems and accreditation quickly became a concern for many laboratories in the EPPO region. EPPO Standards have been approved on quality assurance providing specific guidance for plant pest diagnostics (how to perform the validation of tests, how to organize interlaboratory comparisons...). Standards are regularly revised to update them with most recently developed approaches in accreditation and validation. Laboratories in the EPPO region are increasingly working under quality assurance systems and the need for laboratories to have access to well characterised biological reference material for morphological identification and for the use, development and validation of tests has been recognized. EPPO participated in an EU FP7 project on collections (Q-collect) which resulted in the development of a white paper on collections. Finally, to increase active collaboration among the Organizations involved in plant health research activities at national and regional levels, Euphresco (European Phytosanitary Research Coordination) was established to favour synergies among national research activities and to support plant health policy. Euphresco members identify research priorities to be tackled through transnational collaboration. Many research projects have been funded including projects to develop new tests, validate these tests or evaluate the proficiency of laboratories. These approaches and the experiences in the EPPO region will be presented.

Hébrard Eugénie, Galzi Agnès, Oludare A., Poulicard Nils, Aribi Jamel, Fabre S., Issaka S., Mariac Cédric, Dereeper Alexis, Albar Laurence, Silue D., Fargette D. J. (2018)

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